Liquid Culture Protocol

Name(s):

Part being worked on:
Bacteria:
Antibiotic used: Time and Date:

I. If using an agar plate with transformed bacteria…

1. Look for isolated colonies on the agar plate.
2. On the cover of the plate circle, date, and initial the colony that you are going to transfer to the liquid culture.
3. Sterilize an inoculating loop submerging it in ethanol and briefly putting it in the flame of a Bunsen burner. Let the loop cool for several seconds so the heat doesn't kill the bacteria.
4. Gently touch the inoculating loop to the colony that you are going to transfer and place the loop into a tube of LB broth with the correct antibiotic. Twirl the loop in the tube.
   Antibiotic used:
   Amount of antibiotic used:
   Concentration of antibiotic:
5. Loosely tape the unscrewed caps of the liquid culture to the top of the tube so that the opening is mostly covered.
6. Incubate the bacteria in the tube on a shaker plate at 225 rpm overnight at 37° Celsius.

II. If taking the bacteria directly from the glycerol stock…

1. Take a sterile inoculating loop and scrape it on the glycerol stock to get your bacteria.
   a. A small amount of stock will suffice.
2. Put the inoculating loop in a tube containing LB broth and the correct antibiotic.
   b. The amount of antibiotic should be calculated so that the correct concentration of antibiotic is met.
      Antibiotic used:
      Amount of antibiotic used:
      Concentration of antibiotic:
3. Incubate the bacteria in the tube on a shaker plate at 225 rpm overnight at 37 degrees Celsius.

Location of the incubating bacteria:

Next step: